

SURVIVAL OF THE OYSTER *CRASSOSTREA VIRGINICA*
(GMELIN) IN THE LABORATORY UNDER THE EFFECTS
OF OIL DRILLING FLUIDS SPILLED IN THE LAGUNA DE
TAMIAHUA, MÉXICO

by
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ABSTRACT

In 1965, 970.12 m³ of oil drilling fluid were spilled in the Laguna de Tamiahua, Mexico. Laboratory experiments were carried out to determine possible effects of this upon the oyster *Crassostrea virginica*. It was found that drilling fluid reduced the survival of oysters to a significant degree in concentrations above 200 ppm. At turbidities between 200 and 500 ppm, there was 50% survival on the seventh day. *Tanino* in concentrations between 90 and 170 ppm had a drastic effect upon survival which was 50% between the fourth and fifth days. *Bentonita* in 110 to 190 ppm resulted in 50% survival on the eighth day. *Barita* in concentrations between 50 and 65 ppm did not produce noxious effects on the survival of the oysters. Natural mud in concentrations from 200 to 500 ppm was favorable for the survival of oysters.

INTRODUCTION

With the appearance and increase of internal combustion engines, which increased the demand for oil production, the problem of marine pollution became more pronounced (Yee 1967).

This problem occurs in marine waters, coastal lagoons and other aquatic media as well, but when a fisheries resource is involved, the problem acquires importance beyond the purely biological fields. Yee (*op. cit.*) presented selected references on pollution of marine waters as the result of oil drilling and related activities. His work includes articles written since 1950, in many of which the ramifications of this problem are discussed.

Sugimoto *et al.* (1964, 1965) pointed out that on the fishing grounds of the Seto Inland Sea, damage to the fisheries increased with increasing oil pollution.

The mortality of oysters in relation to natural environments and to oil fields has been analyzed by Mackin and Hopkins

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(1962) in Louisiana. Daugherty (1950, 1951) reported experimental results of the effect of some chemicals used in oil well drilling on marine animals, including the oyster (*C. virginica*); this author explains the reason for his work as follows: "With the recent increase of oil wells in Texas bays, the possibility of pollution from chemical compounds used in drilling became important. Exact knowledge of the effect of these compounds on marine organisms was needed."

The magnitude of the problem is in contrast to the amount of information available, as previous works relating to the effect of fluids from oil drilling on animal life, including the oyster, are very few and deal only with adults. Nothing has been published to date concerning the larval stages of oysters, to which the damaging effects of strange elements in the environment could be even more important.

Very little is known of the relationship that may exist between the turbidity produced by muds, normally found around oyster beds in the Laguna de Tamiahua, and herein called natural muds, and the survival of oysters under similar conditions as mentioned also by Mackin and Hopkins (*op. cit.*).

In Mexico we have had occasion to watch several spills, fortunately most of them without drastic biological results, of oil drilling fluids into Tamiahua Lagoon, Veracruz, one of the most important oyster producing localities of Mexico (fig. 1). In this lagoon, 970.12 m³ of oil drilling fluids were spilled between April and December 1965; this material was composed of 314.79 m³ of material extracted from the different geological strata and 655.33 m³ of industrial materials introduced during the drilling (*vide* Villalobos *et al.* 1968, mimeographed).

This investigation was undertaken to clarify the alleged "mortality" and "extermination" of the oyster reefs in Tamiahua Lagoon. The oil company was sued for a considerable sum of money as recompense for the alleged damage. However, the claim has never been proven by any evidence. The Institute of Biology of the National Autonomous University of Mexico was asked to undertake research on this matter and to give an expert opinion. This was included in the work cited (Villalobos *et al.*) along with some of the data in the present article.

The purpose of this article is to report some results of experiments conducted under laboratory conditions to determine the effects of (a) oil drilling fluids used by Pemex in Tamiahua Lagoon; of (b) several compounds used in the drilling fluids; and of (c) natural mud, on the survival of the local oyster.

ACKNOWLEDGEMENTS

The author wishes to thank Dr. Alejandro Villalobos-Figueroa, leader of the project, whose direction and support were

the principal factors in the progress of the project; and Dr. Gordon Gunter, Director of the Gulf Coast Research Laboratory, Ocean Springs, Mississippi, USA, who kindly joined us in camp at Cucharas and in the Laboratory in Mexico City during September 1967 and whose advice and constructive recommendations were appreciated. It must be noted that Doctor Gunter feels that the experiment reported here should be repeated until the results are more satisfactorily proven. The author is in agreement, but considering the lack of economic resources needed, wished to make these incomplete results available to persons interested in this matter. Dr. Sammy M. Ray was kind enough to provide us with copies of the articles of F. M. Daugherty cited herein, and with other interesting information as well.

Many of my co-workers and friends helped in the construction of aquaria in Cucharas, and also in many other ways; special thanks are hereby expressed to the following: Guadalupe de la Lanza, Fernando Manrique, Samuel Gómez, Andrés Reséndez, Virgilio Arenas, Alberto Ramírez, Gerardo Green and Luis Soto.

Many of the residents of Cucharas and La Laja, Veracruz, were of great help in the every day course of our experimental work, for which I am also very grateful. Drs. A. Villalobos and G. Gunter reviewed and criticized this article. Dr. Allan Phillips assisted in the preparation of the English version.

I also wish to thank: ingenieros Graciano Bello, Guillermo Bernal, Enrique Noguera, Ignacio Cervantes and their collaborators, for their extensive assistance in the field work. The experimental work was made possible by the economic support of Petroleos Mexicanos (PEMEX), under contract to the Instituto de Biología, Universidad Nacional Autónoma de México.

MATERIAL AND METHODS

The oysters used in the experiments came from the Laguna de Tamiahua, particularly from the oyster reefs known as follows: Restinga de Cucharas, La Martinica and Boqueron de Burros (fig. 1). These organisms belong taxonomically to *Crassostrea virginica* (Gmelin), and their medium size was 8 cm in length, with a range from 7 to 12 cm. These oysters were collected with "gafas," an instrument composed of two wooden rakes, 3 meters long, joined a third of the way up, the teeth being nails set in two lines, one opposite the other. These instruments are modified oyster tongs made of wood and nails. Oysters were kept under laboratory conditions for 3 to 5 days before being used in the experiments.

A working place was improvised in the Pemex camp, in the village of Cucharas (fig. 1). A system with a capacity to supply

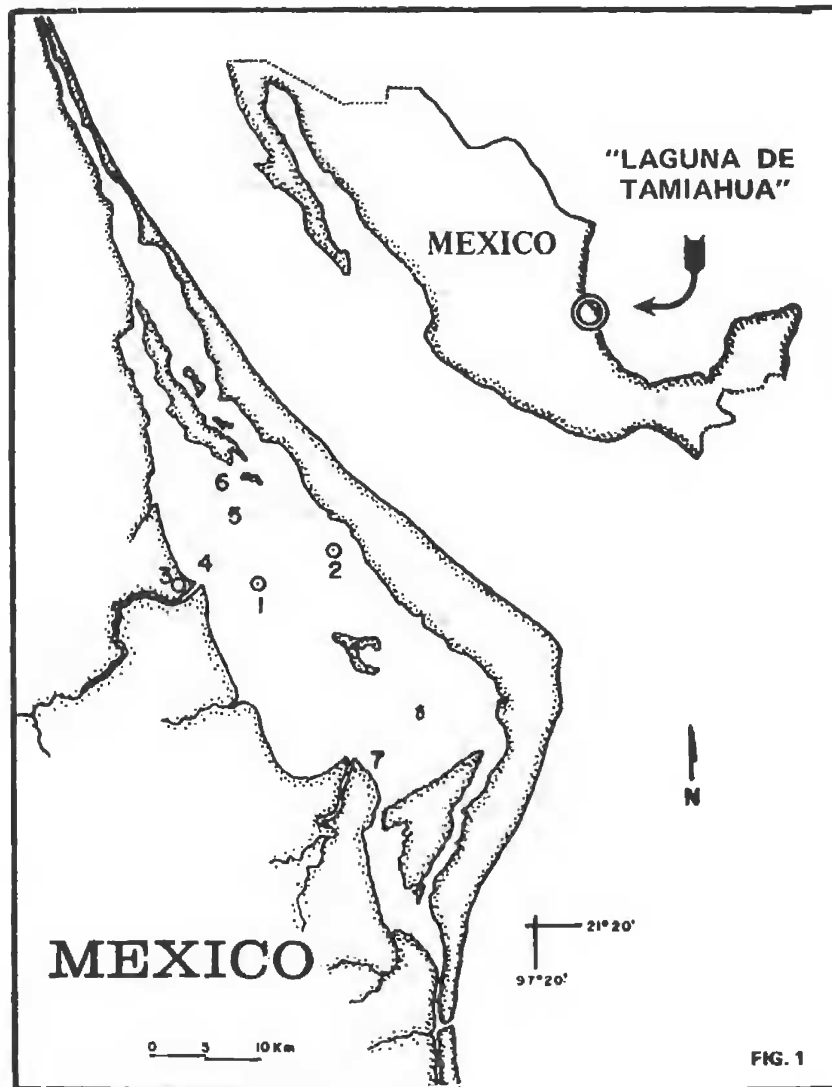


Fig. 1. *Laguna de Tamiahua*:

- 1) *Catán* oil well
- 2) *Acamaya* oil well
- 3) Village and estuary of *Cucharas*
- 4) *Restinga de Cucharas* (oyster reef)
- 5) *Restinga la Martinica* (oyster reef)
- 6) *Boquerón de Burros* (oyster reef)
- 7) *Campanario* (mud test site)

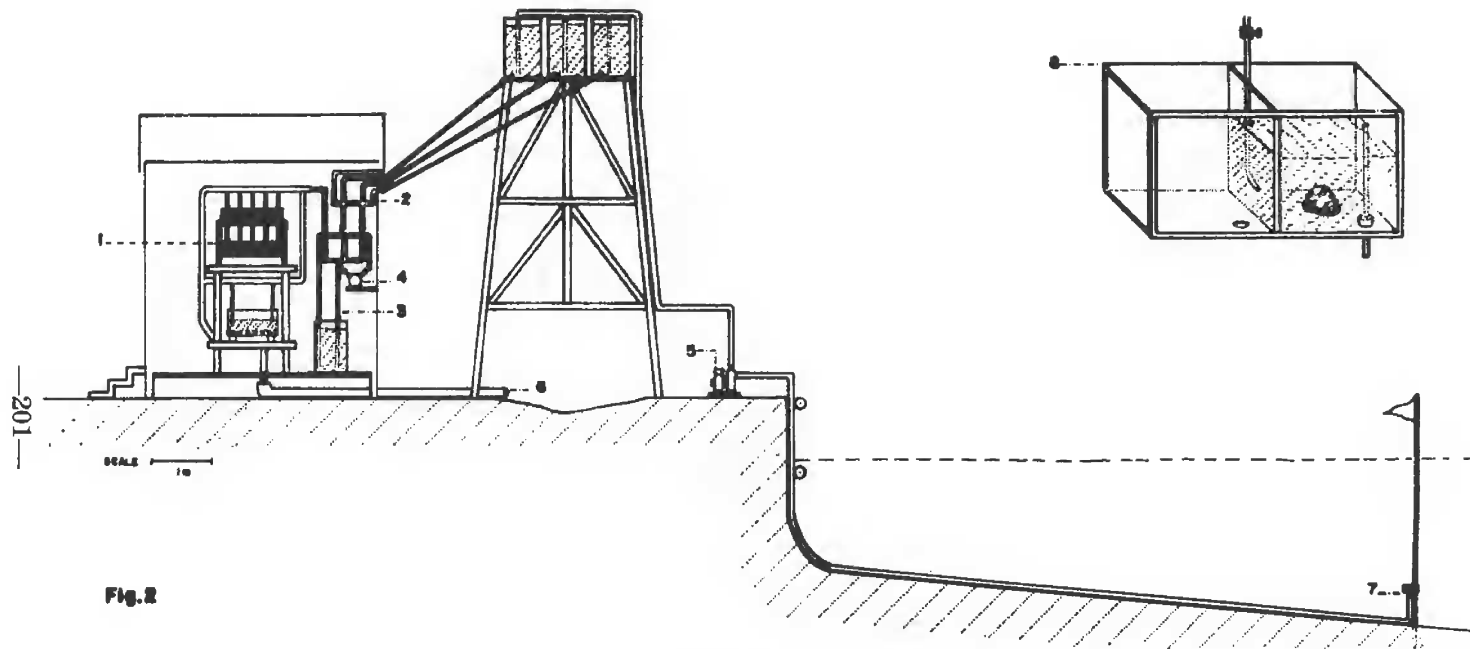


Fig. 2

Fig. 2. Diagram of the system used to supply water and suspended material to the aquaria:

- | | |
|--|---|
| 1) Aquaria | 5) Pump to raise water to the upper tanks |
| 2) Plastic tubing | 6) Outlet |
| 3) Suction tube for suspended material | 7) Intake |
| 4) Plastic pump to inject suspended material | 8) Aquaria with one oyster each |

80 aquaria, each containing 8 liters, was built (fig. 2). Proportions of the aquaria were 20 x 20 x 20 cm, in units of two, back-to-back (fig. 2). Each one had a glass excess drainage tube to regulate the water level. Wooden tables covered with water-proof resin were made as well as supports for the aquaria. The whole system was installed in a wooden cottage.

The water used in the experiments came from the Estero de Cucharas (fig. 1), at a point where a little pier extended some 10 meters from the shore in front of the encampment. It was taken at 40 to 60 cm above the bottom in water of 2 or 2.5 meters deep (fig. 2). The water was raised by "Sentinel" pump, model C 1½ A, powered by a Briggs and Stratton 4-cycle gasoline engine. The water was stored in three asbestos-cement tanks having total capacity of 600 liters. These tanks were located on top of a metal tower 5 meters high.

Water was then distributed by gravity, using a system of plastic and P.V.C. tubing and valves, to adjust the water flow in each aquarium. Water was changed simultaneously in all the aquaria three times a day, each change taking 15 minutes at a flow of two liters per minute; this was done at intervals of approximately eight hours. At the time of changing the water, either test fluids or muds were added to the system by means of a plastic pump (Desmo Plastic-Tec, S. A.) driven by a ¼ H. P. electric motor (Power Electrica S. A.) There was some difficulty in running electric motors in Cucharas, as (at the time of this work) the village lacked public power, so a small diesel electric plant was used according to our needs.

Ingenieros Melesio Muñoz R. and Héctor Soto Rosiles, of the Drilling Department of Pemex, provided the drilling fluids needed for the experiments. They also provided information on the chemical compositions of the drilling fluids, which we were not in a position to analyze. According to this information, the drilling fluid was composed of two fractions: one a combination of different commercial substances and the other the various materials extracted from the geologic strata. The fluid used in the experiments came from a drilling located close to the Laguna de Tamiahua, and made at the same time that the oyster survival experiments were run.

The two above-mentioned engineers were of the opinion that the geologic structure of this drilling was quite similar to that found in the Laguna de Tamiahua; that there were no important differences in the drilling procedures; and that the drilling fluids used in the oyster survival experiments were very similar quantitatively and qualitatively to those spilled in the Laguna de Tamiahua.

The following list of materials introduced in the drilling in the Laguna de Tamiahua, and in what proportions, was pro-

vided by the personnel of Pemex (the names of the chemicals are those known in Mexico, with their sources).

	Kg.	%
Barita	6000	6.5
(Industria Mexicana, S. A., Av. Madero 16, despacho 305, México, D. F.)		
Bentonita	20300	22.0
(Industria Mexicana, S. A.)		
Pirofosfato tetrasódico	1330	1.4
(Hooker Mexicana, S. A., Apartado Postal 7529, México 1, D. F.)		
Tanino Cabel	815	0.8
(Productos Cabel, S. A., Génova 39-105, Mexico 6, D. F.)		
Tinex	200	0.2
(Oleoquímica Monterrey, S. A., Montana 13, 7° Piso, México 18, D. F.)		
C.M.C.	240	0.2
(Deribados Macroquímicos, S. A., Durango 283, México, D. F.)		
Obturante #8	350	0.3
(Productora y Abastecedora, S. A., Apartado Postal 19-512, México, D. F.)		
Lubrisesa	25	0.1
(Sosa Escamas, S. A., Apartado Postal 45, Santa Clara, Estado de México)		
Cromato de Sodio	120	0.1
(Dow Chemical, México, D. F.)		
Diesel	59500	64.7
(PEMEX Mexico)		
Cemento Portland ...	3000	3.2
(México)		

Turbidity was estimated by the method and with the instruments of Jackson, using a turbidimeter 75 cm long. After each change of water in the aquaria, samples were taken for the measurements of turbidity, which was estimated twice in each sample. The maximum of turbidity decreased because of both the sedimentation of suspended material itself and the capacity of oysters to subtract suspended material from the water.

The aquaria were cleaned once a day by emptying them and scouring their walls and floor.

Chlorinity was estimated according to the method of Mohr-Knudsen, dissolved oxygen by the Winkler method, and temperature was taken with a Celsius thermometer. The frequency of these estimations was varied according to the conditions of each experiment; the minimum frequency was once every other day.

Nine experiments are considered in this article: two of a preliminary nature; three to estimate the effect of drilling fluids; three more to show the effect of some components of such drilling fluids in certain concentrations; and one experiment using natural mud.

In most of these experiments survival was observed in two samples—an experimental and a control sample—each containing 20 oysters.

One oyster was placed in each aquarium; a total of 320 oysters was used in these experiments. The experiments were run between March and August 1967, during periods when I had the opportunity to stay in the encampment.

RESULTS

In figures 3 to 11 the results of the experiments are represented graphically. Figure 12 shows the index of 50% of oyster mortality in various concentrations of drilling fluids. Table 1 presents information on the variation of the environment in the aquaria.

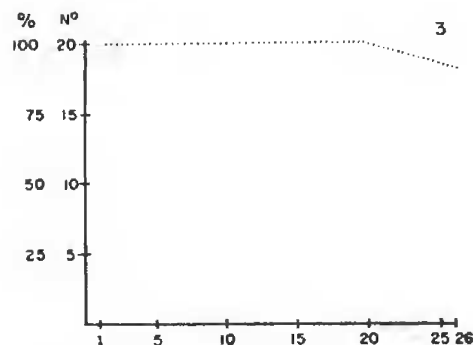


Fig. 3. After 26 days, during which the water was changed three times a day, 85% of the oysters had survived, showing a high index of survival. The ordinate shows the percentage and number of survivors; the abscissa the number of days tested.

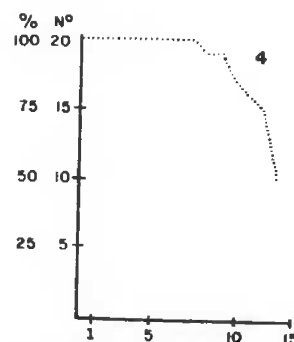


Fig. 4. Mortality was high when water was not changed; 50% survival was reached after 14 days. In the ordinate, survival; in the abscissa, time.

Preliminary experiments. A first experiment was conducted to obtain information on oyster survival under the optimum conditions of changing water which could be maintained for a long time; this would give an index of high survival. A second experiment, without changing the water, was set up to get an index of low survival, in contrast with the first.

After 26 days of the first experiment, during which the water was changed three times a day as mentioned above, 85% of the oysters had survived (fig. 3). This percentage was considered a sufficiently high index of survival, so this frequency of changing water was judged adequate for running the subsequent experiments.

The second experiment shows that 50% mortality was reached after 14 days (fig. 4); this was considered a low index of survival, as it showed that mortality could be high if the water were not changed.

Effects of the drilling fluid. Three experiments were established to test the effects of three different ranges of turbidity and of the drilling fluid.

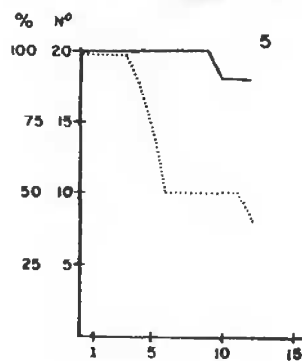


Fig. 5

Survival of two samples, the experimental (dotted line) and the control (continuous line). The first one was treated three times a day with drilling fluid up to 1000 to 2000 ppm of initial turbidity; 50% mortality was reached on the sixth day. In the ordinate, survival; in the abscissa, time.

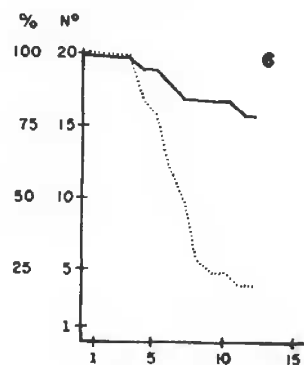


Fig. 6

Survival of two samples, the experimental (dotted line) and the control (continuous line). The first one treated three times a day with drilling fluid in initial turbidity between 200 and 500 ppm. 50% mortality occurred on the seventh day. In the ordinate, survival; in the abscissa, time.

The substances were used in the same form in which they are sold commercially. The concentration of each was arbitrary, always starting from the same volume of dry material suspended or dissolved in a given volume of water. As the properties of each substance are different in regard to solubility and suspensibility, the resulting turbidity was different for each component.

Tanino in turbidity between 90 and 170 ppm: 50% mortality was reached between the fourth and the fifth day in the experimental sample; mortality was total on the seventh day. The controls only reached 20% mortality on the seventh day (fig. 8).

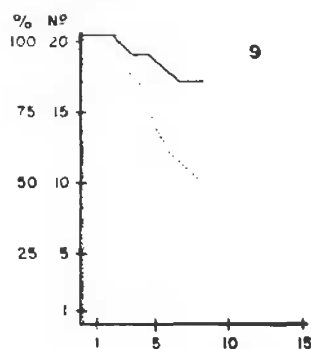


Fig. 9.

Survival of two samples, the experimental (dotted line) and the control (continuous line). The first one treated three times a day with Bentonita in initial turbidity between 110 and 190 ppm. 50% survival occurred on the eighth day. In the ordinate, survival; in the abscissa, time.

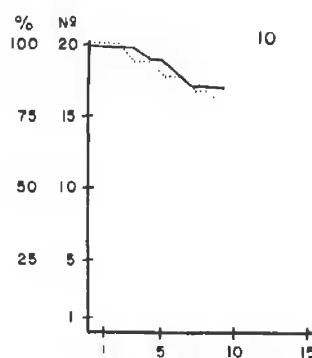


Fig. 10.

Survival of two samples, the experimental (dotted line) and the control (continuous line). The first one treated three times a day with Barita in initial turbidity between 50 and 65 ppm. Both survival curves were quite similar, so that no lethal effect was shown. In the ordinate, survival; in the abscissa, time.

Bentonita in concentrations between 110 and 190 ppm: 50% survival in the experiment sample occurred on the eighth day; at this time the control lot reached 20% mortality (fig. 9).

Barita in concentrations between 50 and 65 ppm: on the ninth day, survival was 80% in the experimental sample and 85% in the control. The survival curves were quite similar, so that no lethal effect was shown (fig. 10).

Daugherty (1951) reported some results on the action of Tanino, called in his paper "tannex"; he established that one lot of 24 oysters survived 22.5 hours in increased concentrations up to 140 ppm in a system with recirculating water and constant turbidity, and also found that a similar lot survived 20.5 hours in increased concentrations up to 450 ppm. From this the author concluded that tannex was not toxic to oysters; however, he proved that it is toxic to other marine organisms in concentrations of 70 to 450 ppm over the same period of time. Apparently the results of Daugherty and those presented in this article are not in agreement, but what caused the difference is probably the methodology. The main factor may be the length of time the experiment was run; however, the concentrations used and their variations, and the frequency of changing the water in the aquaria, also may be significant factors. In my opinion, Daugherty's experiments did not last long enough to show any mortality of oysters.

The Bentonita in 110 to 190 ppm proved to cause significant mortality in the experiment here reported; but in the opinion of the experts of Pemex this substance forms compounds of high density in the drilling fluid, so that its suspensibility is not great and it could not be scattered over a broad area in sufficient concentration at one time, to produce a significant mortality in the Laguna de Tamiahua. This opinion is supported and discussed also by Villalobos *et al.* (1968) referring to the total drilling fluid.

Daugherty (1951) reported that the "aquagel," a trade name for a high quality Bentonita, did not kill any oysters during 22 hours in concentrations as high as 7500 ppm. This author considered "aquagel" as non-toxic to marine animals. As in the case of the Tanino, the results obtained in the present experiments are apparently in disagreement with Daugherty's. The same major objection applies again: Daugherty's did not last long enough to kill the animals.

The Barita in the concentrations used in this experiment did not produce any apparent mortality as can be seen from the survival curves (fig. 10). In this respect Daugherty (1951) using "baroyd," made of selected Barita, also found this substance to be non-toxic either to the oyster or to the other marine animals of his experiments. Our findings are in agreement despite differences in methodology.

It is interesting to add that Daugherty (1950) found "sodium acid pyrophosphate" to be toxic to oysters in concentrations of 500 ppm and greater. This substance was not tested in our experiments.

In my opinion none of the information reported by Daugherty (1950-1951) or such of the present articles as concerns

the effect of components of the drilling fluid, could be applicable to the case of the Laguna de Tamiahua, as none of these substances was spilled in its commercial form; rather, they were used as components of the drilling fluid which was then used in the drilling, and it was only at the end of the drilling operations that the fluid was spilled. For these reasons it is assumed that the only information available that could be applicable to the case of the Laguna de Tamiahua is that presented herein referring to the effect of the drilling fluid upon the oysters. The applicability of this information, however, is not so obvious, and even it may be of doubtful value for many reasons. The most important of these reasons is that the laboratory experiments are not a complete reproduction of what happened at the time when the drilling fluids were spilled. Furthermore these experiments deal only with oysters and drilling fluid, without taking into consideration the whole ecosystem of which the oysters were a component; the drilling fluid, as a foreign substance, probably affected the usual functioning of the ecosystem, but at present no information is available on this difficult and complex problem. What is most desirable is to avoid the spilling of any foreign substance into coastal lagoons such as Tamiahua, without limiting the activities of the oil industry and other industries. This despite the opinion of Daugherty (1951) that the compounds tested, for him, were sufficiently low in toxicity to be of little danger when released in open bay waters.

As to the effect of natural mud on the survival of oysters, the high survival of the experimental lot in contrast with the controls, seems to show that this substance favors survival. Mackin and Hopkins (1962) pointed out that in certain localities in Louisiana, natural mortality of oysters was in inverse proportion of the water's turbidity; in places and periods with high turbidity, the mortality was lower. These results are in agreement with mine, but the reasons for this effect are not clear.

During the experiments variations were recorded in temperature, chlorinity and the concentrations of dissolved oxygen. This information did not require special treatment so only the limits were reported (Table 1). Temperature varied from 23° to 30°C, as a result of seasonal variations from spring to summer. Chlorinity varied between 4.40 and 16.57‰. Lower values were found during a short period in August in relation to the rainy season in summertime.

The concentrations of dissolved oxygen varied between 3.0 and 7.6 ml/L; it was highest during the intake of water. Low values of oxygen were found only at the end of the second experiment, probably due to fermentation and oxidation in the stagnant water. It seems logical to believe that temperature,

chlorinity and dissolved oxygen fluctuated between narrow limits so as to give comparable results in the various experiments.

Availability of food was not controlled, but it seems that oysters can survive long periods without abundant food, according to experiments conducted by Dr. Sammy M. Ray (personal communication).

CONCLUSIONS

Drilling fluid reduced the survival of oysters significantly in concentrations of over 200 ppm in laboratory aquaria. In turbidities between 200 and 500 ppm, 50% mortality was reached on the seventh day. According to Villalobos *et al.* (1968), these concentrations could hardly be maintained in time and space sufficient to produce significant mortality or the claimed extermination of the oyster reefs in the Laguna de Tamiahua.

This conclusion is the only one that could be applicable to the case of the Laguna de Tamiahua, as all the others deal with commercial components of the drilling fluid, and these were not spilled as components. Nothing is known about the effect of drilling fluid upon the ecosystem to which oysters belong.

Tanino in concentrations between 90 and 170 ppm had a drastic effect upon survival, which was 50% between the fourth and fifth days.

Bentonita in 110 to 190 ppm resulted in 50% survival on the eighth day.

Barita in concentrations between 50 and 65 ppm did not produce noxious effects on the survival of the oysters.

Natural mud in concentrations from 200 to 500 ppm was favorable for the survival of oysters.

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A BIBLIOGRAPHY OF ANOMALIES
OF FISHES

Supplement 2

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This listing adds 213 titles to the original bibliography (Gulf Res. Repts. 1(6), 1964) and the 1966 supplement (Gulf Res. Repts. 2(2):169-176). Although a number of previously overlooked references are included, there are 137 citations of works published during the 1965-70 period. The frequency of recent publications offers some indication of the current worldwide interest in problems of fish teratology.

Apparently in response to increased interest in the environment, some American authors are now attempting to relate anomalies in natural or "wild" populations of fishes to pollution levels. While this may be a useful area of investigation, the results should be evaluated with caution. It is obvious that only sub-lethal conditions are observed and there are few data, especially in marine or estuarine environments, upon which to establish a "normal" survival potential for a particular abnormality in any species. Those hoping to use published records to establish a "base-line" should bear in mind that, with few exceptions, both old and recent publications report only the most obvious and striking conditions (macro-anomalies). Investigators attempting to determine frequencies of abnormalities should include data on the less obvious and possibly very common micro-anomalies.

This supplement includes an Index of Sources and Abbreviations covering those journals not included in the original bibliography. As in the previous supplement, titles are serially numbered and prefixed by the letter "S"; the prefix has been omitted from the index listings. Corrections to be made in the original bibliography are indicated under Corrigenda.

I again wish to express my continuing appreciation to those colleagues who have provided reprints of their papers or otherwise contributed to this bibliography.

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